

WHAT IS CLAIMED IS:

1. A non-human transgenic animal whose germ cells and somatic cells contain a knockout mutation in DNA encoding 4E-BP1, and wherein said transgenic
5 animal shows a phenotype of an altered glucose and/or fat metabolism as compared to a control animal.

2. The transgenic animal of claim 1, wherein said animal is a mammal.

10 3. The mammal of claim 2, wherein said mammal is a mouse.

4. A cell line derived from the non-human transgenic animal of claim 1.

15 5. A cell line derived from the mouse of claim 3.

6. A method of producing a non-human transgenic animal, in which at least some cells thereof contain an altered gene encoding an altered 4E-BP1, said altered gene having been targeted to disrupt the wild type 4E-BP1 gene in said transgenic animal, said method comprising:

20 a) providing an altered gene encoding the altered form of 4E-BP1 and designed to target and disrupt said wild type 4E-BP1 gene of an embryonic stem cells (ES) of said animal;

b) introducing said altered gene in said ES cells;

25 c) selecting ES cells in which said altered 4E-BP1 gene has disrupted said wild type 4E-BP1 gene;

d) injecting said selected ES cells of c) into blastocysts;

e) implanting said blastocysts of d) in a pseudopregnant animal; and

30 f) producing a transgenic animal having at least some cells having said altered 4E-BP1 gene encoding said altered 4E-P1.

7. A method of identifying an agent which modulates glucose or fat metabolism *in vivo* comprising:

a) administering an agent suspected of being a modulator of cap-dependent translation in an animal;

5 b) measuring glucose and/or lipid levels in the animal of step a) and comparing same with that of a control animal, not having been administered said agent, wherein a difference in glucose and/or lipid levels of the animal of step a) as compared to that of the control animal identifies said agent as a modulator of glucose or fat metabolism *in vivo*.

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8. The method of claim 7, wherein said positive modulator of cap-dependent translation is a modulator of the level or of the activity of 4E-BP1.

9. The method of claim 8, wherein said agent increases and/or strengthens
15 the interaction of 4E-BP1 with eIF-4E.

10. The method of claim 7, wherein said agent decreases and/or weakens the interaction of 4E-BP1 with eIF-4E.

20 11. The method of claim 7, wherein said agent decreases the amount of eIF-4F pre-initiation complex, thereby decreasing the translation of mRNAs implicated in glucose or lipid metabolism.

12. The method of claim 11, wherein said decrease in the amount of eIF-4F pre-initiation complex involves a sequestration of eIF-4E in a complex with an eIF-4E
25 sequestering agent.

sub 2 13. ~~The method of claim 12, wherein eIF-4E-sequestering agent comprises a sequence having an amino acid sequence selected from YxxxL ϕ , Yx+xf $\phi\phi$, + ϕ xYx+xf $\phi\phi$, + $\phi\phi$ Y-xf/A $\phi\phi$ xxRxSP, and + $\phi\phi$ Y-xfL ϕ xxRxSP, or + ϕ xYx+xfL ϕ xxxxxx~~

wherein + and - refer to a charged amino acid; ϕ is a hydrophobic amino acid; x is any amino acid; and the capital letters refer to the known one letter code for amino acids

14. The method of claim 13, wherein said sequestering agent is selected from
5 4E-BP, eIF4G, p82, p150, p130 and p20.

15. The method of claim 14, wherein said sequestering agent is selected from
4E-BP1, 4E-BP2, 4E-BP3, eIF-4G1, eIF4-G2, 4E-BP, and eIF-4G.

10 16. The method of claim 15, wherein said sequestering agent is selected from
4E-BP1, 4E-BP2, 4E-BP3, eIF-4G1, eIF-4G2, 4E-BP, and eIF-4G.

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sub 2³ 17. Method of identifying an agent which modulates glucose and/or fat
metabolism *in vivo* comprising:
15 a) providing a translationally active preparation of translation factors
and at least one mRNA having a cap structure whose translation is cap-dependent;
b) measuring the initiation of translation on said mRNA, or the
binding of at least some translation factors of a) to said cap of said mRNA in the
presence and in the absence of an agent suspected of modulating the translation
20 efficiency of cap-dependent mRNAs or the binding of translation factors to the cap
structure thereof, thereby identifying an agent which modulates cap-dependent translation
and wherein a difference in the translation activity and/or binding in the presence of the
agent, as compared to that in the absence thereof identifies said agent as a modulator of
cap-dependent translation;
25 c) administering said agent identified in b) to an animal; and
d) measuring glucose and/or lipid levels in the animal of step c) and
comparing same with that of a control animal, not having been administered said agent,
wherein a difference in glucose and/or lipid levels of the animal of step c) as compared
to that of the control animal identifies said agent as a modulator of glucose or fat
30 metabolism *in vivo*.

18. The method of claim 17, where the agent is obtained from a library of compounds.

19. The method of claim 18, wherein the animal is a mammal.

20. The method of claim 19, wherein said mammal is a mouse or human.

sub 24 21. A modulator of glucose or fat metabolism ~~in vivo identified by any one of the methods of claims 17, 18, 19 or 20.~~

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22. A method of decreasing fat tissue growth and/or weight gain, comprising:

a) administering an agent which desequsters eIF-4E from a sequestering agent, thereby increasing the amount of eIF-4E available for a formation of eIF-4F preinitiation complex, leading to an increase of translation of cap-dependent mRNAs implicated in a reduction of a tissue growth and/or weight gain.

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23. The method of claim 22, wherein said sequestration of eIF-4E is through its interaction with 4E-BP1.

24. The method of claim 23, wherein said desequestration of eIF-4E from 4E-BP1 is effected by an antibody specific to the eIF-4E interaction domain of 4E-BP1, or an epitope-bearing portion thereof.

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25. The method of claim 24, wherein said desequestration or said inhibition of said sequestration is effected by an inhibition of the synthesis of 4E-BP1.

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26. The method of claim 25, comprising an agent which inhibits the synthesis of 4E-BP1, wherein said agent comprises an antisense RNA complementary to the nucleotide sequence encoding for 4E-BP1.

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27. A method of determining whether an agent modulates fat tissue growth and/or weight gain in an animal comprising:

- a) providing a translationally active preparation of translation factors and at least one mRNA having a cap structure whose translation is cap-dependent;
- 5 b) measuring the initiation of translation on said mRNA, or the binding of at least some translation factors of a) to said cap of said mRNA in the presence and in the absence of an agent suspected of modulating the translation efficiency of cap-dependent mRNAs or the binding of translation factors to the cap structure thereof, thereby identifying an agent which modulates cap-dependent translation
- 10 and wherein a difference in the translation activity and/or binding in the presence of the agent, as compared to that in the absence thereof identifies said agent as a modulator of cap-dependent translation;
- c) administering said agent identified in b) to an animal; and
- d) measuring fat tissue growth and/or weight gain in the animal of
- 15 step c) and comparing same with that of a control animal, not having been administered said agent, wherein a difference in fat tissue growth and/or weight gain of the animal of step c) as compared to that of the control animal identifies said agent as a modulator of fat tissue growth and/or weight gain *in vivo*.

20 28. The method of claim 27, where the agent is obtained from a library of compounds.

29. The method of claim 28, wherein the animal is a mammal.

25 30. The method of claim 29, wherein said mammal is a mouse or human.

sub 25 31. A modulator of glucose or fat metabolism *in vivo* identified by any one of the methods of claims 28, 29, 30 or 31.

32. A method of treating obesity, comprising administering to an obese animal, or an animal susceptible of becoming obese, an agent which increases the amount of eIF-4E available for a formation of eIF-4F preinitiation complex.

5 33. The method of claim 32, wherein said agent is an agent which
desequesters eIF-4E from 4E-BP1.

34. A method of determining whether an agent modulates obesity in an animal comprising:

10 a) providing a translationally active preparation of translation factors and at least one mRNA having a cap structure whose translation is cap-dependent;

b) measuring the initiation of translation on said mRNA, or the binding of at least some translation factors of a) to said cap of said mRNA in the presence and in the absence of an agent suspected of modulating the translation efficiency of cap-dependent mRNAs or the binding of translation factors to the cap structure thereof, thereby identifying an agent which modulates cap-dependent translation and wherein a difference in the translation activity and/or binding in the presence of the agent, as compared to that in the absence thereof identifies said agent as a modulator of cap-dependent translation;

cap-dependent translation;

20 c) administering said agent identified in b) to an animal; and

 d) assessing obesity in the animal of step c) and comparing same

with that of a control animal, not having been administered said agent, wherein a

difference in obesity of the animal of step c) as compared to that of the control animal

identifies said agent as a modulator of obesity *in vivo*.

25 35. The method of claim 18, where the agent is obtained from a library of
compounds.

36. The method of claim 35, wherein the animal is a mammal.

30 37. The method of claim 36, wherein said mammal is a mouse or human.

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38. A modulator of glucose and/or fat metabolism ~~in vivo~~ identified by any one of the methods of claims 7, 17, 22, 27, 34, 35 or 36.

39. A method of treating diabetes type II and associated complications,
5 comprising administering to an animal suffering from type II diabetes, or at risk of suffering therefrom, an agent which increases the amount of eIF-4E available for a formation of eIF-4F preinitiation complex.

40. The method of claim 39, wherein said agent is an agent which
10 desequesters eIF-4E from 4E-BP1.

41. A method of identifying an agent which modulates glucose and/or fat metabolism *in vivo* comprising:

- 15 a) incubating a portion of eIF-4E capable of directly binding with a peptide comprising an eIF-4E-binding domain;
- b) assessing said direct binding between said portion of eIF-4E and said peptide in a presence and in an absence of an agent wherein an agent which potentially modulates glucose and/or fat metabolism is identified when a difference in said binding in the presence of said agent, as compared to that in the absence thereof is
20 observed;
- c) administering said agent identified in b) to an animal; and
- d) measuring glucose and/or lipid levels in the animal of step c) and comparing same with that of a control animal, not having been administered said agent, wherein a difference in glucose and/or lipid levels of the animal of step c) as compared
25 to that of the control animal identifies said agent as a modulator of glucose or fat metabolism *in vivo*.

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